PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Seroprevalence of SARS-CoV-2 antibodies in children of healthcare workers- A prospective multicentre cohort study protocol
AUTHORS	Corr, Michael; Christie, Sharon; Watson, Chris; Maney, Julieann; Fairley, Derek; Ladhani, Shamez N.; Lyttle, Mark; McFetridge, Lisa; Mitchell, Hannah; Shields, Michael; McGinn, Claire; McKenna, James; Mallett, Peter; Ferris, Kathryn; Rowe-Setz, Gala; Moore, Rebecca; Foster, Steven; Evans, Jennifer; Waterfield, Tom

VERSION 1 – REVIEW

REVIEWER	yadong Gao Wuhan University, China.
REVIEW RETURNED	30-Jun-2020

GENERAL COMMENTS	The paper is well written and clearly described the protocol of the ongoing study of seroprevalence of SARS-CoV-2 antibody in children. I have no any concern about the ethics, design, sample size and statistic of this protocol.

REVIEWER	Roger Zemek and Maala Bhatt Children's Hospital of Eastern Ontario, Canada Dr. Bhatt and Dr. Zemek are co-PIs on study examining SARS-CoV-2 antibody levels in households funded by the province of Ontario (Canada).
REVIEW RETURNED	30-Jul-2020

GENERAL COMMENTS	The Seroprevalence of SARS-CoV-2 Antibodies in Children – A Prospective Multicentre Cohort Study Thank you for the opportunity to review this study protocol. The authors are conducting a multicenter cohort study at five centres in the United Kingdom to determine the prevalence of antibodies to SARS-CoV-2 in children of healthcare workers. They are aiming to recruit a convenience sample of 1000 children to detect a 10% change in prevalence at each site at each sampling time point. The investigators have a parent/patient advisory board which is a significant strength. As the study is currently recruiting, the comments that follow are suggestions in light of the study status. GENERAL COMMENTS 1. Given our evolving knowledge of the shorter than expected duration of SARS-CoV-2-specific antibodies, the investigators could consider conducting a sample size calculation based on

	precision of estimates rather than change in prevalence of antibodies. We expect that antibodies might drop off 3-months post-infection. 2. In order to expand the conclusions from this study, reporting results from household testing particularly the healthcare worker parent should be considered. Information such as molecular testing in the healthcare worker parent and the result, symptoms consistent with SARS-CoV-2 in parents if molecular testing was negative and molecular testing in other members of the household and the result could be appended to the data collection. 3. The authors acknowledge the limitation in generalizability of their study given that they will only be recruiting children of healthcare workers. Since they will be recruiting participants using advertisements and social media they could consider recruiting a second cohort of children from the general population to provide insight on the difference in prevalence between these populations. SPECIFIC COMMENTS 1. Suggest changing the title to “Seroprevalence of SARS-CoV-2 Antibodies in Children of Health Care Workers – A prospective multicenter cohort study” to more accurately reflect the study population. 2. Consider using an assay that is able to measure IgM and IgG titers (as compared to a dichotomous yes/no). 3. Determining neutralizing antibodies is mentioned as an objective but is not included in the secondary outcomes and there is no information provided regarding the proposed tests for neutralizing antibodies. 4. There is insufficient information provided about the timing of molecular testing and distribution of symptom questionnaires. Table 1 indicates that testing will be done when symptomatic, but no further information is provided in the protocol. The study visits occur at baseline, 2- and 6-months. What happens if a participant is symptomatic between visits? 5. How will the study team be made aware that testing has been done or convey to the participant should have a test completed if symptomatic? 6. How will the study team distribute the symptom questionnaires to the participants? 7. The symptom questionnaire currently includes some medical jargon (e.g., lethargy, photophobia). Not all healthcare workers necessarily have a medical vocabulary; consider revising the survey language to ensure reading comprehension for all potential participants. 8. Trial registration and amendment dates are not consistent with the dates on clinicaltrials.gov. Please revise.

REVIEWER	Kirk Tickell University of Washington
REVIEW RETURNED	31-Jul-2020

GENERAL COMMENTS	The paper is well written and the longitudinal follow-up will greatly improve in the impact of the study. The authors correctly acknowledge that recruiting children of healthcare workers is likely to be biased, but that the data will still valuable. However, there are some information that the authors should provide more clarity and depth in:

	Major issues 1) There is no description of how you will recruit the children of healthcare workers – are you approaching them at work, phoning them, leaving flyers on wards. It is really important you clarify the step between selecting a hospital and the actual enrollment. 2) In the informed consent section, it is mentioned that consent will be obtained from parents, but for older children will you obtain a assent or consent if they are caperable from them? Will you recruit a 13-year old child whose mother says yes, but the child refuses to be involved – presumably not. 3) In the abstract you say the age range is 2-15 years, in the article summary you say older 6 months. 4) You refer to healthcare workers a lot – it would be good to define who is healthcare work, do hospital cleaners count? What cadres are you referring to. 5) For the symptomatic diary, how do you define "onset of illness", this seem very subjective. Will caregivers only record onset of symptoms they think might be COVID? Please clarify the instructions they will be given about which symptoms to report. 6) It would be to clarify what will happen if a child tests positive, presumably they will be picked up by Public Health authorities, and their caregiver will be tested. Is there a concern caregivers may choose to enroll, or not, based on their own exposure to COVID patients (e.g. I think I might have been exposed so I want to child tested, or I don't want my child tested because we might both quarantined?) Will there be a method to track the number of eligible healthcare workers vs those that choose to enroll? Minor issues 7) In the abstract you say the age range is 2-15 years, in the article summary you say older 6 months. 8) In the statistical plan you say "adjusted probabilities" (Line 41) – what are they adjusted for. Are you a-priori adjusting deciding to age-adjust them, or are you saying you'll look at a list of risk factors and use a rule to decide which are confounders.

VERSION 1 – AUTHOR RESPONSE

Reviewer #1:

Thank you for your positive review.

Reviewer #2:
Thank you for a very informative and helpful peer review.
GENERAL COMMENTS
1. Given our evolving knowledge of the shorter than expected duration of SARS-CoV-2-specific antibodies, the investigators could consider conducting a sample size calculation based on precision of estimates rather than change in prevalence of antibodies. We expect that antibodies might drop off 3-months post-infection.

We discussed this within the team and the feel that because the study has been funded and received favourable research ethics committee reviews based on the power calculations presented we cannot change them.

2. In order to expand the conclusions from this study, reporting results from household testing particularly the healthcare worker parent should be considered. Information such as molecular testing in the healthcare worker parent and the result, symptoms consistent with SARS-CoV-2 in parents if molecular testing was negative and molecular testing in other members of the household and the result could be appended to the data collection.

We agree entirely with this suggestion and have already introduced additional data collection in keeping with this using RedCap. We have also included copies of the CRF in the supplement and have included the following text in the methods:

“Study data will be collected and managed using REDCap (Research Electronic Data Capture) electronic data capture tools (22). Participants and their parents will information relating to illness episodes, suspected household exposure to SARS-CoV-2 and the outcome of any molecular testing. These data will be collected at each clinic visit.”

3. The authors acknowledge the limitation in generalizability of their study given that they will only be recruiting children of healthcare workers. Since they will be recruiting participants using advertisements and social media they could consider recruiting a second cohort of children from the general population to provide insight on the difference in prevalence between these populations.

This is an excellent idea. We had similar thoughts and have included a comparison group of non-patient facing hospital staff such as managers and secretarial staff. We hope that by including this group we can make the results more generalizable.

SPECIFIC COMMENTS
1. Suggest changing the title to “Seroprevalence of SARS-CoV-2 Antibodies in Children of Health Care Workers – A prospective multicenter cohort study” to more accurately reflect the study population.

This has been changed as advised

2. Consider using an assay that is able to measure IgM and IgG titers (as compared to a dichotomous yes/no).

We were exploring assays at the time of writing. We now have agreement to test using ROCHE, Abbott and Diasorin assays – These will provide titre results and we intend to report titre results in addition to a binary positive/negative based on the manufacturers suggested cut-offs.

3. Determining neutralizing antibodies is mentioned as an objective but is not included in the secondary outcomes and there is no information provided regarding the proposed tests for neutralizing antibodies.

This is a key point – thank you. We are not actually testing if the antibodies are neutralising but rather testing for antibodies to the spike protein (likely to be neutralising). We have therefore removed all reference to neutralising antibodies and instead focussed on the assays themselves.

4. There is insufficient information provided about the timing of molecular testing and distribution of

symptom questionnaires. Table 1 indicates that testing will be done when symptomatic, but no further information is provided in the protocol. The study visits occur at baseline, 2- and 6-months. What happens if a participant is symptomatic between visits?

Thank you. We had debated as to the best approach. As reviewer 3 eludes to any testing needs to be available to the Public Health Agencies also. Thankfully at all sites routine testing is available for all children as part of the Public Health response. We have therefore agreed that testing can be completed using routine services that feed into Public Health. The participants will then provide the result to the study team. The symptom diaries and CRF data will be managed electronically using RedCap. Additional details added in the text and in Table 1.

5. How will the study team be made aware that testing has been done or convey to the participant should have a test completed if symptomatic?

Testing will be conducted as part of the existing Public Health response. Participants to feed results back to researchers.

6. How will the study team distribute the symptom questionnaires to the participants?

This has been made clearer and the following text has been added.

“Study data will be collected and managed using REDCap (Research Electronic Data Capture) electronic data capture tools (22). Participants and their parents will provide information relating to illness episodes, suspected household exposure to SARS-CoV-2 and the outcome of any molecular testing at each clinic appointment. Participants that test positive for SARS-CoV-2 via real-time RT-QPCR will be required to complete a symptom diary from the onset of their illness until resolution. Participants will also be provided with electronic symptom diaries to record any illness episodes relating to possible COVID-19 contemporaneously. In all instances symptoms of illness episodes will be recorded prior to antibody test results being disclosed to minimise recall bias. (Copy of symptom diaries and case report forms are available in the online supplement).”

7. The symptom questionnaire currently includes some medical jargon (e.g., lethargy, photophobia). Not all healthcare workers necessarily have a medical vocabulary; consider revising the survey language to ensure reading comprehension for all potential participants.

Thank you, we suspect an old version of the symptom diary had been attached. The current version includes explanations of both lethargy (sleeping more than usual) and photophobia (dislike of lights). This version has been attached now.

8. Trial registration and amendment dates are not consistent with the dates on clinicaltrials.gov. Please revise.

Corrected

Reviewer #3

Thank you for your detailed peer review.

Major issues
1) There is no description of how you will recruit the children of healthcare workers – are you

approaching them at work, phoning them, leaving flyers on wards. It is really important you clarify the step between selecting a hospital and the actual enrollment.

This has been clarified in the text. We approached healthcare workers via internal intranet advertisements and email circulars. The following text has been added:

“Participants will be recruited from each participating NHS organisation using internal intranet advertisements and email circulars.”

2) In the informed consent section, it is mentioned that consent will be obtained from parents, but for older children will you obtain a assent or consent if they are caperable from them? Will you recruit a 13-year old child whose mother says yes, but the child refuses to be involved – presumably not.

This has been clarified to include assent for children under 16 with the following text added:

“Informed consent will be obtained prior to inclusion including assent from the child. The parent/child is free to decline/withdraw consent at any time without providing a reason and without being subject to any resulting detriment. For children who turn 16 during the follow up period they will be invited to consent for the study again. If the young person declines consent they will be withdrawn from the study without being subject to any resulting detriment. Additional consent will be sought to store specimens for future research”

3) In the abstract you say the age range is 2-15 years, in the article summary you say older 6 months.

This has been corrected

4) You refer to healthcare workers a lot – it would be good to define who is healthcare work, do hospital cleaners count? What cadres are you referring to.

Healthcare workers has been defined as follows:

“Children of healthcare workers who are aged between 2 and 15 years old. For the purpose of this study, a healthcare worker is defined as an employee of the National Health Service. Healthcare workers will be categorized based on their role and if that role involves patient facing activities or not. A group of approximately 200 non-clinical and non-patient facing staff such as managerial staff and secretaries will be included to provide a comparison to clinical staff and improve the generalizability of the results. Participants will be recruited from each participating NHS organisation using internal intranet advertisements and email circulars.”

We have opted to include non-clinical staff as per reviewer 2 to provide greater generalizability of results.

5) For the symptomatic diary, how do you define “onset of illness”, this seem very subjective. Will caregivers only record onset of symptoms they think might be COVID? Please clarify the instructions they will be given about which symptoms to report.

The participants will be asked to provide any symptoms they feel may be due to COVID-19. Copies of the symptom diary and RedCap data collection tools have been added and the following text has been added also

“Participants will be asked to record from their perceived first day of illness until their perceived last day of illness. “

6) It would be to clarify what will happen if a child tests positive, presumably they will be picked up by Public Health authorities, and their caregiver will be tested. Is there a concern caregivers may choose to enroll, or not, based on their own exposure to COVID patients (e.g. I think I might have been exposed so I want to child tested, or I don't want my child tested because we might both quarantined?) Will there be a method to track the number of eligible healthcare workers vs those that choose to enroll?

This is a key point. Thankfully at all sites molecular testing is available for all symptomatic children via the Public Health pandemic response. To ensure we comply with rules around reporting we have asked participants to arrange testing via the routine mechanism and feedback results to the study team. Details have been added. This will also reduce the risks as suggested around a need to quarantine or not.

Minor issues

7) In the abstract you say the age range is 2-15 years, in the article summary you say older 6 months.

This has been corrected.

8) In the statistical plan you say "adjusted probabilities" (Line 41) – what are they adjusted for. Are you a-priori adjusting deciding to age-adjust them, or are you saying you'll look at a list of risk factors and use a rule to decide which are confounders.

This has been added- Logistic regression will be used to estimate the probability of the presence of SARS-CoV-2 infection, adjusting for factors including demographic features (age, gender) and symptomology (such as fever, cough, fatigue), that are deemed statistically significant.

VERSION 2 – REVIEW

REVIEWER	Kirk Tickell University of Washington
REVIEW RETURNED	20-Aug-2020

GENERAL COMMENTS	The authors have addressed my concerns.